

**WHAT IS CLAIMED IS:**

1. An apparatus for measuring cellular electrical conditions comprising a Cell Support Membrane component adapted to hold one or more cells which includes: i) a first layer comprising a non-  
5 conductive material comprising a top surface and bottom surface and including one or more pores, wherein the top surface of the material comprises one or more cell attachment sites which circumscribe the pores of the material and contact the cells, and wherein the pores of the material are capable of forming electrically tight seals with the contacted cells at the  
10 cell attachment sites, and ii) a second layer comprising a non-porous, non-conductive, sealant material which contacts the first layer of the Cell Support Membrane.

2. The apparatus according to claim 1, wherein the cellular electrical conditions are selected from the group consisting of  
15 transmembrane potential, capacitance, resistance, and conductance.

3. The apparatus according to claim 1, wherein the first layer of the Cell Support Membrane component comprises material selected from the group consisting of glass, plastic, rubber, polytetrafluorotethylene, polytetrafluorotethylene/glass, polyethylene terephthalate, and  
20 polycarbonate.

4. The apparatus according to claim 1, wherein the second layer of the Cell Support Membrane component comprises material selected from the group consisting of polytetrafluorotethylene, polytetrafluorotethylene/glass, polyethylene terephthalate, and  
25 polycarbonate.

5. The apparatus according to claim 1, wherein the second layer of the Cell Support Membrane component comprises material selected from the group consisting of polyhydroxybutyrate, polylactate, polyglycolic acid, polycaprolactone, cellulose, starch, and collagen.

5                    6. The apparatus according to claim 1, wherein the second layer of the Cell Support Membrane component comprises a dye.

7. The apparatus according to claim 6, wherein the second layer of the Cell Support Membrane component comprises Solvent Blue 14.

10                    8. The apparatus according to claim 1, wherein the cell attachment sites of the first layer of the Cell Support Membrane component are treated with a composition comprising molecules that facilitate cell attachment.

15                    9. The apparatus according to claim 8, wherein the molecules are selected from the group consisting of gelatin, poly-L-lysine, poly-D-lysine, collagen, and fibronectin.

20                    10. The apparatus according to claim 1, wherein an area of the first layer of the Cell Support Membrane component outside of the cell attachment sites is treated with a composition comprising molecules that inhibit cell attachment.

11. The apparatus according to claim 10, wherein the molecules are selected from the group consisting of silane<sup>7</sup>, silicone, and Teflon®.

12. The apparatus according to claim 1, wherein the first layer of the Cell Support Membrane component displays at least 4 pores.

13. The apparatus according to claim 1, wherein the first layer of the Cell Support Membrane component displays 1 pore.

5                    14. The apparatus according to claim 1, wherein the pores of the first layer of the Cell Support Membrane component are between 0.2  $\mu\text{m}$  and 2  $\mu\text{m}$  in diameter.

10                    15. The apparatus according to claim 1, wherein the cells are selected from the group consisting of HEK-293 cells, Chinese hamster ovary cells, primary neuronal cells, skeletal muscle cells, smooth muscle cells, cardiac muscle cells, immune cells, epithelial cells, and endothelial cells.

15                    16. The apparatus according to claim 15, wherein the primary neuronal cells are selected from the group consisting of hippocampus, dorsal root ganglia, and superior cervical ganglia cells.

20                    17. The apparatus according to claim 1, wherein the cells comprise DNA constructs directing the expression of ~~molecules~~ selected from the group consisting of ion channel proteins, ion transporters, G-proteins, G-protein ligands, G-protein modulators, G-protein receptors, protein kinases, and protein phosphatases.

18. The apparatus according to claim 1, wherein the cells express ion channels that are specific for ions selected from the group consisting of sodium, potassium, calcium, and chloride.

19. The apparatus according to claim 1, wherein the cells are permeabilized by contact with any one of the following: i) an antibiotic selected from the group consisting of amphotericin and nystatin; ii) a detergent selected from the group consisting of digitonin and saponin; or iii) a high voltage field.

20. The apparatus according to claim 1, wherein the second layer is positioned under the first layer of the Cell Support Membrane component, and an area of the second layer of the Cell Support Membrane component that is in contact with a pore that is contacted with a cell is selectively removed.

21. The apparatus according to claim 20, wherein the area of the second layer of the Cell Support Membrane component is removed by microscope-assisted photo-ablation.

22. The apparatus according to claim 21, wherein the microscope is a confocal microscope.

23. The apparatus according to claim 21, wherein the photo-ablation is carried out with a flash lamp.

24. The apparatus according to claim 21, wherein the photo-ablation is carried out with a laser.

25. The apparatus according to claim 24, wherein the laser is selected from the group consisting of argon, helium/neon, krypton, YAG, and titanium-sapphire laser.

26. The apparatus according to claim 1, wherein the second layer is positioned over the first layer of the Cell Support Membrane component, and an area of the second layer of the Cell Support Membrane component that is in contact with a pore that is contacted with a cell is selectively removed.

27. The apparatus according to claim 26, wherein the area of the second layer of the Cell Support Membrane component is removed by an enzyme selected from the group consisting of proteases, cellulases, esterases, and depolymerases, which is secreted by the cell.

28. The apparatus according to claim 1, further comprising a chamber to hold the Cell Support Membrane component which includes a top area and bottom area, wherein the cell attachment sites of the Cell Support Membrane component face the top area of the chamber.

29. The apparatus according to claim 28, further comprising electrolyte solution which contacts the first and second layers of the Cell Support Membrane component.

30. The apparatus according to claim 29, further comprising two electrodes, Electrode 1 and Electrode 2, that are placed in the electrolyte solution, wherein one Electrode 1 is a 'ground' electrode and Electrode 2 is a current-passing/voltage-measuring electrode, and wherein one of the two electrodes faces the top surface of the first layer of the Cell Support Membrane component, and one of the two electrode faces the bottom surface of the first layer of the Cell Support Membrane component.

31. The apparatus according to claim 30, further comprising local pre-amplification circuitry, and a voltage-clamp, current-clamp and lock-in amplifier.

32. An apparatus for measuring cellular electrical  
5 conditions comprising a Cell Support Membrane component adapted to hold one or more cells which comprises a non-conductive material that includes a top surface and bottom surface and includes one or more pores, wherein:  
i) the top surface of the material comprises one or more cell attachment sites which circumscribe the pores of the material and contact the cells; ii)  
10 the pores of the material are capable of forming electrically tight seals with the contacted cells at the cell attachment sites, and iii) a pore that is not sealed with a cell is plugged by the addition of one composition to the top surface of the material, and another composition to the bottom surface of the material, such that the compositions interact in the pore and form a non-  
15 conductive solid product.

33. The apparatus according to claim 32, wherein the cellular electrical conditions are selected from the group consisting of transmembrane potential, capacitance, resistance, and conductance.

34. The apparatus according to claim 32, wherein the one  
20 composition comprises an enzyme and the other composition comprises a substrate for the enzyme.

35. The apparatus according to claim 32, wherein the one composition comprises calf alkaline phosphatase and the other composition comprises 5-bromo-4-chloro-3'-indolyphosphate p-toluidine salt and nitro-  
25 blue tetrazolium.

36. The apparatus according to claim 32, wherein the material comprising the Cell Support Membrane component is selected from the group consisting of glass, plastic, rubber, polytetrafluoroethylene, polytetrafluoroethylene/glass, polyethylene terephthalate, and polycarbonate.

37. The apparatus according to claim 32, wherein the cell attachment sites of the Cell Support Membrane component are treated with a composition comprising molecules that facilitate cell attachment.

38. The apparatus according to claim 37, wherein the molecules are selected from the group consisting of gelatin, poly-L-lysine, poly-D-lysine, collagen, and fibronectin.

39. The apparatus according to claim 32, wherein an area outside of the cell attachment site is treated with a composition comprising molecules that inhibit cell attachment.

40. The apparatus according to claim 39, wherein the molecules are selected from the group consisting of silane, silicone, and Teflon®.

41. The apparatus according to claim 32, wherein the Cell Support Membrane component displays at least 4 pores.

42. The apparatus according to claim 32, wherein the Cell Support Membrane component displays 1 pore.

43. The apparatus according to claim 32, wherein the pores are between 0.2  $\mu\text{m}$  and 2  $\mu\text{m}$  in diameter.

44. The apparatus according to claim 32, wherein the cells are selected from the group consisting of HEK-293 cells, Chinese hamster ovary cells, primary neuronal cells, skeletal muscle cells, smooth muscle cells, cardiac muscle cells, immune cells, epithelial cells, and endothelial cells.

45. The apparatus according to claim 44, wherein the primary neuronal cells are selected from the group consisting of hippocampus, dorsal root ganglia, and superior cervical ganglia cells.

46. The apparatus according to claim 32, wherein the cells comprise DNA constructs directing the expression of molecules selected from the group consisting of ion channel proteins, ion transporters, G-proteins, G-protein ligands, G-protein modulators, G-protein receptors, membrane receptors, protein kinases, and protein phosphatases.

47. The apparatus according to claim 32, wherein the cells express ion channels that are specific for ions selected from the group consisting of sodium, potassium, calcium, and chloride.

48. The apparatus according to claim 32, wherein the cells are permeabilized by contact with any one of the following: i) an antibiotic selected from the group consisting of amphotericin and nystatin; ii) a detergent selected from the group consisting of digitonin and saponin; or iii) a high voltage field.

49. The apparatus according to claim 32, further comprising a chamber to hold the Cell Support Membrane component which includes a top area and bottom area, wherein the Cell Support Membrane



component is positioned in the chamber so that the cell attachment sites of the Cell Support Membrane face the top area of the chamber.

50. The apparatus according to claim 49, further comprising electrolyte solution which contacts the first and second layers of the Cell Support Membrane component.

51. The apparatus according to claim 50, further comprising two electrodes, Electrode 1 and Electrode 2, that are placed in the electrolyte solution, wherein one Electrode 1 is a 'ground' electrode and Electrode 2 is a current-passing/voltage-measuring electrode, and wherein one of the two electrodes faces the top surface of the first layer of the Cell Support Membrane component, and one of the two electrode faces the bottom surface of the first layer of the Cell Support Membrane component.

52. The apparatus according to claim 51, further comprising pre-amplification circuitry and a voltage-clamp, current-clamp and lock-in amplifier.

53. An apparatus for measuring cellular electrical conditions comprising a Cell Support Membrane component adapted to hold cells which comprises a non-conductive material including a top surface and bottom surface and including one or more pores, wherein: i) the top surface of the material comprises one or more cell attachment sites which circumscribe the pores of the material and contact the cells; ii) the pores of the material are capable of forming electrically tight seals with the contacted cells at the cell attachment sites, and iii) the cells are directed to the pores by an attractant.

54. The apparatus according to claim 53, wherein the cellular electrical conditions are selected from the group consisting of transmembrane potential, capacitance, resistance, and conductance.

55. The apparatus according to claim 53, wherein the  
5 attractant is a chemoattractant.

56. The apparatus according to claim 55, wherein the chemoattractant is selected from the group consisting of growth factors, chemokines, and nutrients.

57. The apparatus according to claim 56, wherein the  
10 chemoattractant is 20-hydroxyleukotrine B4.

58. The apparatus according to claim 53, wherein the attractant is an electric field.

59. The apparatus according to claim 53, wherein the cells are resuspended in a density gradient.

60. The apparatus according to claim 59, wherein the  
15 density gradient comprises suspensions selected from the group consisting of percoll, ficoll, and metrizamide.

61. The apparatus according to claim 53, wherein the material comprising the Cell Support Membrane component is selected from  
20 the group consisting of glass, plastic, rubber, polytetrafluorotethylene, polytetrafluorotethylene/glass, polyethylene terephthalate, and polycarbonate.

62. The apparatus according to claim 53, wherein the cell attachment sites of the Cell Support Membrane component are treated with a composition comprising molecules that facilitate cell attachment.

5 63. The apparatus according to claim 62, wherein the molecules are selected from the group consisting of gelatin, poly-L-lysine, poly-D-lysine, collagen, and fibronectin.

64. The apparatus according to claim 53, wherein an area outside of the cell attachment sites is treated with a composition comprising molecules that inhibit cell attachment.

10 65. The apparatus according to claim 64, wherein the molecules are selected from the group consisting of silane, silicone, and Teflon®.

66. The apparatus according to claim 53, wherein the Cell Support Membrane component displays at least 4 pores.

15 67. The apparatus according to claim 53, wherein the Cell Support Membrane component displays 1 pore.

68. The apparatus according to claim 53, wherein the pores are between 0.2  $\mu\text{m}$  and 2  $\mu\text{m}$  in diameter.

20 69. The apparatus according to claim 53, wherein the cells are selected from the group consisting of HEK-293 cells, Chinese hamster ovary cells, primary neuronal cells, skeletal muscle cells, smooth muscle cells, cardiac muscle cells, immune cells, epithelial cells, and endothelial cells.

70. The apparatus according to claim 69, wherein the primary neuronal cells are selected from the group consisting of hippocampus, dorsal root ganglia, and superior cervical ganglia cells.

71. The apparatus according to claim ~~53~~, wherein the cells  
5 comprise DNA constructs directing the expression of molecules selected from the group consisting of ion channel proteins, ion transporters, G-proteins, G-protein ligands, G-protein modulators, G-protein receptors, membrane receptors, protein kinases, and protein phosphatases.

72. The apparatus according to claim 53, wherein the cells  
10 express ion channels that are specific for ions selected from the group consisting of sodium, potassium, calcium, and chloride.

73. The apparatus according to claim 53, wherein the cells  
are permeabilized by contact with any one of the following: i) an antibiotic selected from the group consisting of amphotericin and nystatin; ii) a  
15 detergent selected from the group consisting of digitonin and saponin; or iii) a high voltage field.

74. The apparatus according to claim 53, further comprising a chamber to hold the Cell Support Membrane component which includes a top area and bottom area, wherein the Cell Support Membrane  
20 component is positioned in the chamber so that the cell attachment sites of the Cell Support Membrane face the top area of the chamber.

75. The apparatus according to claim 74, further comprising electrolyte solution placed in the chamber to contact the first and second layers of the Cell Support Membrane component.

76. The apparatus according to claim 75, further comprising two electrodes, Electrode 1 and Electrode 2, that are placed in the electrolyte solution, wherein one Electrode 1 is a 'ground' electrode and Electrode 2 is a current-passing/voltage-measuring electrode, and wherein  
5 one of the two electrodes faces the top surface of the first layer of the Cell Support Membrane component, and one of the two electrode faces the bottom surface of the first layer of the Cell Support Membrane component.

77. The apparatus according to claim 76, further comprising pre-amplification circuitry and a voltage-clamp, current-clamp  
10 and lock-in amplifier.

78. An apparatus for measuring cellular electrical conditions comprising a Microchip component adapted to hold cells which comprises non-conductive material including a top surface and bottom surface, wherein the top surface of the material includes cell attachment  
15 sites that are sized to contact individual cells and are coupled to an electrode lead/signal modifying circuitry.

79. The apparatus according to claim 78, wherein the cellular electrical conditions are selected from the group consisting of transmembrane potential, capacitance, resistance, and conductance.

80. The apparatus according to claim 78, wherein the material of the Microarray component is selected from the group consisting of glass, silicone, plastic, rubber, polytetrafluorotethylene, polytetrafluorotethylene/glass, polyethylene terephthalate, and polycarbonate.

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81. The apparatus according to claim 78, wherein the cell attachment sites are treated with a composition comprising molecules that facilitate cell attachment.

5 82. The apparatus according to claim 81, wherein the molecules are selected from the group consisting of gelatin, poly-L-lysine, poly-D-lysine, collagen, and fibronectin.

83. The apparatus according to claim 78, wherein an area surrounding the cell attachment sites is treated ~~with~~ a composition comprising molecules that inhibit cell attachment.

10 84. The apparatus according to claim 83, wherein the molecules are selected from the group consisting of silane, silicone, and Teflon®.

15 85. The apparatus according to claim 78, wherein the cell attachment sites include pins that are sized to penetrate the contacting cell, and wherein each pin houses an electrode connected to a headstage preamplifier.

86. The apparatus according to claim 78, wherein the cell attachment sites include pits that are sized to accommodate the contacting cells.

20 87. The apparatus according to claim 78, wherein the top surface of the Microchip component displays at least 9 cell attachment sites.

88. The apparatus according to claim 78, wherein the cell attachment sites are between 15  $\mu\text{m}$  and 20  $\mu\text{m}$  in diameter.

89. The apparatus according to claim 78, wherein the cells are selected from the group consisting of HEK-293 cells, Chinese hamster ovary cells, primary neuronal cells, skeletal muscle cells, smooth muscle cells, cardiac muscle cells, immune cells, epithelial cells, and endothelial cells.

90. The apparatus according to claim 89, wherein the primary neuronal cells are selected from the group consisting of hippocampus, dorsal root ganglia, and superior cervical ganglia cells.

91. The apparatus according to claim 78, wherein the cells comprise DNA constructs directing the expression of molecules selected from the group consisting of ion channel proteins, ion transporters, G-proteins, G-protein ligands, G-protein modulators, G-protein receptors, membrane receptors, protein kinases, and protein phosphatases.

92. The apparatus according to claim 78, wherein the cells express ion channels that are specific for ions selected from the group consisting of sodium, potassium, calcium, and chloride.

93. The apparatus according to claim 78, wherein the cells are permeabilized by contact with a high voltage field.

94. The apparatus according to claim 78, wherein a cell attachment site is adjacent to an independent ground electrode and connected to an independent measuring electrode.

95. The apparatus according to claim 78, further comprising a chamber to hold the Cell Support Membrane component which includes a top area and bottom area, wherein the Microchip component is positioned in the chamber so that the cell attachment sites of the Microchip  
5 face the top area of the chamber.

96. The apparatus according to claim 95, further comprising electrolyte solution which contacts the top surface of the Microchip component.

97. The apparatus according to claim 96, further  
10 comprising local pre-amplification circuitry and a voltage-clamp, current-clamp, and lock-in amplifier.

98. An high throughput screening method for detecting and assaying test agents that affect cellular electrical activity comprising: i) attaching cells to a plurality of apparatuses according to claim 31; ii)  
15 measuring electrical activity of the cells contacted with electrolyte solution; iii) contacting the cells with an electrolyte solution comprising a test agent; iv) measuring the electrical activity of the cells contacted with the test agent; and v) assessing a difference between the measured electrical activity in step (ii) and the measured electrical activity in step (iv).

20 99. The method according to claim 98, wherein the cellular electrical activity is selected from the group consisting of potential, resistance, conductance, and capacitance.

100. The method according to claim 98, wherein chambers of the apparatuses are provided by a multi-well plate.



101. The method according to claim 98, wherein the multi-well plate is processed by computer-controlled equipment.

102. The method according to claim 98, wherein the cells are selected from the group consisting of HEK-293 cells, Chinese hamster ovary cells, primary neuronal cells, skeletal muscle cells, smooth muscle cells, cardiac muscle cells, immune cells, epithelial cells, and endothelial cells.

103. The method according to claim 102, wherein the primary neuronal cells are selected from the group consisting of hippocampus, dorsal root ganglia, and superior cervical ganglia cells.

104. The method according to claim 98, wherein the cells comprise DNA constructs directing the expression of molecules selected from the group consisting of ion channel proteins, ion transporters, G-proteins, G-protein ligands, G-protein modulators, G-protein receptors, membrane receptors, protein kinases, and protein phosphatases.

105. The method according to claim 98, wherein the cells express ion channels that are specific for ions selected from the group consisting of sodium, potassium, calcium, and chloride.

106. The method according to claim 98, wherein the cells are permeabilized by contact with any one of the following: i) an antibiotic selected from the group consisting of amphotericin and nystatin; ii) a detergent selected from the group consisting of digitonin and saponin; and iii) a high voltage field.

107. The method according to claim 98, wherein the test agent is selected from the group consisting of neurotransmitters, neurotransmitter analogues, enzyme inhibitors, ion channel modulators, G-proteins, G-protein ligands, G-protein modulators, G-protein receptors, transport inhibitors, hormones, peptides, toxins, antibodies, pharmaceutical agents, and chemicals.

108. The method according to claim 98, wherein the test agent is selected from the group consisting of purinergics, cholinergics, serotonergics, dopaminergics, anesthetics, benzodiazepines, barbiturates, steroids, alcohols, metal cations, cannabinoids, cholecystokinins, cytokines, excitatory amino acids, GABAergics, gangliosides, histaminergics, melatonins, neuropeptides, neurotoxins, endothelins, NO compounds, opioids, sigma receptor ligands, somatostatins, tachykinins, angiotensins, bombesins, bradykinins, and prostaglandins.

109. A high throughput screening method for detecting and assaying cell processes that affect cellular electrical activity comprising: i) attaching cells to a plurality of apparatuses according to claim 31; ii) measuring electrical activity of cells that are in a resting state; iii) initiating the cell process; iv) measuring the electrical activity of cells engaged in the process; and v) assessing a difference between the measured electrical activity in step (ii) and the measured electrical activity in step (iv).

110. The method according to claim 109, wherein the cellular electrical activity is selected from the group consisting of potential, resistance, conductance, and capacitance.

111. The method according to claim 109, wherein chambers of the apparatuses are provided by a multi-well plate.

112. The method according to claim 109, wherein the multi-well plate is processed by computer-controlled equipment.

5 113. The method according to claim 109, wherein the cells are selected from the group consisting of HEK-293 cells, Chinese hamster ovary (CHO) cells, primary neuronal cells, skeletal muscle cells, smooth muscle cells, cardiac muscle cells, immune cells; epithelial cells, and endothelial cells.

10 114. The method according to claim 113, wherein the primary neuronal cells are selected from the group consisting of hippocampus, dorsal root ganglia, and superior cervical ganglia cells.

15 115. The method according to claim 109, wherein the cells comprise DNA constructs directing the expression of molecules selected from the group consisting of ion channel proteins, ion transporters, G-protein coupled receptors, membrane receptors, protein kinases, and protein phosphatases.

20 116. The method according to claim 109, wherein the cells express ion channels that are specific for ions selected from the group consisting of sodium, potassium, calcium, and chloride.

117. The method according to claim 109, wherein the cells are permeabilized by contact with any one of the following: i) an antibiotic selected from the group consisting of amphotericin and nystatin; ii) a

detergent selected from the group consisting of digitonin and saponin; and  
iii) a high voltage field

118. The method according to claim 109, wherein the test agent is selected from the group consisting of neurotransmitters, neurotransmitter analogues, enzyme inhibitors, ion channel modulators, G-proteins, G-protein ligands, G-protein modulators, G-protein receptors, transport inhibitors, hormones, peptides, toxins, antibodies, pharmaceutical agents, and chemicals.

119. The method according to claim 109, wherein the test agent is selected from the group consisting of purinergics, cholinergics, serotonergics, dopaminergics, anesthetics, benzodiazepines, barbiturates, steroids, alcohols, metal cations, cannabinoids, cholecystokinins, cytokines, excitatory amino acids, GABAergics, gangliosides, histaminergics, melatonins, neuropeptides, neurotoxins, endothelins, NO compounds, opioids, sigma receptor ligands, somatostatins, tachykinins, angiotensins, bombesins, bradykinins, and prostaglandins.

120. The method according to claim 109, wherein the cell process is selected from the group consisting of cell-cell interaction, cell-cell fusion, viral infection, endocytosis, exocytosis, membrane recycling, and membrane-ligand interaction